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Division of Radiation and Regeneration Control
Department of Cellular Biology

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The final goal of our department is to establish a system to regenerate or facilitate regeneration of damaged cells by various environmental stresses, especially radiation. For this purpose, we are studying the mechanism for dynamic organization of nuclear functions in stress response especially DNA double strand breaks (DSBs). We have already established the UVA laser microirradiation system to induce DSBs in a restricted nuclear region. We are now studying the dynamics of DNA repair proteins and chromatin in response to DNA damage using the live imaging system and the multicolor immunofluorescence technique. Furthermore, we are establishing a system to analyze the *in vivo* biochemical interaction of DNA repair proteins and damaged chromatin.

Dr. Shoji is studying the induction of neurocristopathy syndrome by environmental factors, especially the development of cardiovascular anomalies such as conotruncal anomalies, and creating an effective model that closely simulates the human one. Also, he is collaborating with the School of Medicine of the University of Tokushima (Professor Fukui) to investigate the developmental mechanism of human syndrome induced by an environmental factor. He attended the Teratology Society 45th Annual Meeting in Trade Winds Island Grand Resort, St. Pete Beach, Florida, USA. June 25-30, 2005. and presented a paper entitled “Conotruncal Anomalies and Neurocristopathy Following Maternal Excess Tretinoin Exposure.” He has also been serving as part time lecturer to teach graduate students at Graduate School of Medical Sciences, University of Tokushima.

1. Dynamic organization of nuclear domains in DNA damage response.

Tashiro S.

To examine the dynamics of chromatin and non-chromatin nuclear domains in the regulation of DNA repair, we have developed a laser UV microirradiation system to induce DNA double strand breaks (DSBs) at the restricted areas in cell nuclei. Using this system, we are studying the dynamics of DNA repair proteins and apoptosis related proteins after induction of DSBs. We have already obtained preliminary results suggesting that the dynamics of non-chromatin nuclear domains in the process of DNA repair are regulated according to the function of each nuclear domain. For the further characterization of the dynamic organization of higher order nuclear architecture, we have combined the Fluorescence Recovery after Photobleaching (FRAP) technique with laser-UV-microirradiation. This new method allows us to examine the dynamics of specific proteins in living cells after induction of DSBs.

2. Study of the mechanism to regulate dynamics and localization of chromatin and non-chromatin nuclear domains.

Tashiro, S.

In this study, we are going to analyze the basic higher order nuclear architecture regulating the formation and dynamics of non-chromatin nuclear domains. For this purpose, we will analyze the localization and dynamics of known nuclear domains using newly established imaging techniques including visualization of nuclear function and living cell imaging system. Furthermore, biochemical approach is applied to identify a key molecule for the formation of non-chromatin nuclear domains as a scaffold.

3. Molecule mechanism analysis of MLL gene translocations in AML

Sun, J., Tashiro, S.

Translocations involving the MLL gene are a frequent finding in infant and adult acute leukemia and are also observed in therapy-related leukemia. Mapping of the chromosomal breakpoint has revealed that almost all of the breakpoints cluster within an 8. 3-kb region. To understand the mechanism of translocation in this region, chromatin structure and recruitment of DNA repair proteins are investigated.

4. Function analysis of Bach1 in regulation of heme response gene

Sun, J., Tashiro, S., Igarashi, K.

Though the transcription factors such as NF-E2, GATA-1 are known as regulators of the globin gene, the mechanism of globin gene expression remains explanation. We performed quantitative RT-PCR analysis using bone marrow cells from wild- type and bach1 deficient mice. beta-globin mRNA in bach1-/- mutant was decreased. We also determined the expression of hematopoietic specific transcription factors such as NF-E2 and GATA-1. Their mRNA showed no significant change in the mutant mice. Moreover, after phenylhydrazine treatment, the platelets were increasing in peripheral blood of bach1-/- mutant mice.

5. Animal model of abnormal vasculogenesis and conotruncal anomalies in Neurocristopathy syndrome induced by environmental factors.

Shoji, S.

This study examines through autopsy the genetic effects of numerous anomalies found at various gestational ages in aborted and stillbirth fetuses and in intrauterine death from both the atomic bomb survivors and those who did not experience the bomb. It then investigates, from the developmental biology stand point, the morphogenesis of these anomalies found in various organs, especially in the cardiovascular system, which are experimentally induced using a variety of environmental agents. The results are used in creation of animal model of teratogenic neurocristopathy syndrome that are resembled in human DiGeorge and Velocardiofacial (CATCH 22) syndromes, more specifically the teratogenic anomalies of the cardiovascular system including conotruncal anomalies, in order to clarify their pathogenesis.

6. Radiation or environment agent-induced teratogenesis in the offspring following exposures.

Shoji, S.

The present study was conducted to determine whether following DNA damage induced by ionizing radiation or environmental agents leads to lethality and teratogenesis among the progeny and to compare them to those reported in

humans.

In order to understand the mechanism of embryonic lethality and teratogenesis in the F_1 offspring caused by environmental agents, we continue to study such occurrences among the offspring of parental mice that have been exposed to environmental agents such as radiation and chemical substances.

Even a single abnormal development during fertilization and embryogenesis is known to result in lethality and/or teratogenesis, with DNA damage and individual genetic background having strong influences in these processes. It is considered that these genetic factors are inherited by the next generation, and the study investigates such processes for lethality and teratogenesis.

A. Original Papers

1. Toki T[*1], Katsuoka F[*1], Kanezaki R[*1], Xu G[*1], Kurotaki H[*1], Sun J, Kamio T[*1], Watanabe S[*1], Tandai S[*1], Terui K[*1], Yagihashi S[*1], Komatsu N[*1], Igarashi K[*2], Yamamoto M[*3], Ito E[*1] ([*1]Hirosaki Univ., [*2]Dept. Biomed. Chem., [*3]Tsukuba Univ.) Transgenic expression of Bach1 transcription factor results in megakaryocytic impairment. Blood. 105(8): 3100-8. 2005

2. Fujiwara T[*1], Harigae H[*1], Takahashi S[*1], Furuyama K[*1], Nakajima O[*2], Sun J, Igarashi K[*3],Yamamoto M[*4], Sassa S[*5], Kaku M[*1], Sasaki T[*1]. ([*1]Tohoku Univ., [*2]Ymagata Univ., [*3]Dept. Biomed. Chem., [*4]Tsukuba Univ., [*5]Rockefeller Univ.) Differential gene expression profiling between wild-type and ALAS2-null erythroblasts: identification of novel heme-regulated genes. Biochem Biophys Res Commun. 340(1): 105-10. 2006

3. DohiY[*1], Alam J[*2], Yoshizumi M[*3], Sun J, Igarashi K[*1]. ([*1]Dept. Biomed. Chem., [*2]Louisiana State Univ. [*3]Dept. Cardiovascul. Physiol.) Oxygenase-1 Gene Enhancer Manifests Silencing Activity in a Chromatin Environment Prior to Oxidative Stress. Antioxid Redox Signal. 8(1-2): 60-67. 2006

4. Igarashi K[*], Sun J. ([*]Dept. Biomed. Chem., Tohoku Univ.) The heme-bach1 pathway in the regulation of oxidative stress response and erythroid differentiation. Antioxid Redox Signal. 8(1-2): 107-18. 2006

5. Tashiro S, Cremer M[*], Solovei I[*], Cremer T[*]. ([*]Ludwig Maximilians Univ.) Nuclear architecture: Topology and function of chromatin and non-chroamtin nuclear domains. In “Nuclear dynamics: Approaches from biochemistry, molecular cell biology and visual biology”. Springer Inc. (in press)

B. Meetings

1. Satoshi TASHIRO:Topological analysis of nuclear domains using Multicolor immunofluorescence staining. The 5th Nuclear dynamics meeting, Hakone, 2005.5

2. Satoshi TASHIRO: DNA repair and dynamics of nuclear architecture. The 46th Annual meeting of Research Society for Delayed effect of Atomic Bomb Detonation, Hiroshima, 2005.6.

3. Satoshi TASHIRO, Tsuyoshi IKURA, Kenji KAMIYA: Dynamics of histone H2AX upon DNA damage. The 64th Annual meeting of the Japanese Cancer Association, Sapporo, 2005.9.

4. Satoshi TASHIRO, Kazuki KONO, Yumi HARANO, Akihiko MUTO, Hideto HOSHINO, MInoru YOSHIDA, Kazuhiko IGARARSHI: Regulation of dynamics and function of Bach2 by SUMOylation. The 67th Annual meeting

of Japanese Society of Hematology, Yokohama, 2005. 9.

5. Satoshi Tashiro: Characterization of interchromatin compartment. The 78th Annual Meeting of the Japanese Biochemical Society, Kobe 2005. 10.

6. Tashiro, S.: Characterization of Interchromatin Compartment. International Symposium on Ran and Cell Cycle. Awaji, 2005. 10.

7. Satoshi TASHIRO: Dynamics of nuclear domains associated with DNA repair. 48th Annual Meeting of Japan Radiation Research Society, Hiroshima. 2005. 11.

8. Kazuki KONO, Satoshi TASHIRO, Yumi HARANO, Kazuhiko IGARASHI: Regulation of dynamics and function of Bach2, a transcription repressor, by SUMOylation. The 28th Annual Meeting of the Molecular Biology Society of Japan, Fukuoka, 2005. 12.

9. Satoshi TASHIRO: Topological analysis of nuclear domains using Multicolor immunofluorescence staining. The 23rd Chromosome workshop, Hiroshima, 2006.1.

10. Tashiro, S., Miyagawa, K., Ikura, T., Igarashi, K.: Dynamics of higher order nuclear architecture upon DNA damage. The 3rd International Symposium of Hiroshima University 21st Century COE Program, DNA Damage Response and Cancer. Hiroshima, 2006. 2.

11. Shoji, S., Sawada, K.[1], Shoji, I.[2], Azad MD. AK[1], Fukui, Y.[1] and Tashiro, S. ([1]Department of Anatomy and Developmental Neurobiology, School of Medicine, University of Tokushima, Japan. [2]College of Medicine and Public Health, The Ohio State University, USA) Conotruncal Anomalies and Neurocristopathy Following Maternal Excess Tretinoin Exposure. Teratology Society 45th Annual Meeting at Trade Winds Island Grand Resort, St. Pete Beach, Florida, USA. June 25-30, 2005. (Birth defects res. A: Clinical and Molecular Teratology 73 (5), 361, 2005) (A, R)

12. Shoji, S., Sawada, K.[1], Shoji, I.[2], Azad MD. AK[1], Fukui, Y.[1] and Tashiro, S. ([1]Dept. Anatomy and Developmental Neurobiology, Sch. Med., Univ. Tokushima, [2]College of Medicine and Public Health, The Ohio State University, USA) Conotruncal anomalies and Neurocristopathy following maternal gamma rays and excess Tretinoin exposure. 48th Annual Meeting of the Japan Radiation Research Society / 1st Asian Congress of Radiation Research, Hiroshima, Nov.15-17, 2005. (English Abstracts pp.121; Japanese Abstracts pp.116)